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Alkaline phosphatase as a reporter enzyme.

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This study examines the use of alkaline phosphatase (AP) as a reporter enzyme. We constructed a plasmid containing the cDNA which encodes the bone/liver/kidney rat AP under the control of the simian virus 40 (SV40) early promoter and used it to transfect Chinese hamster ovary, SV40-transformed African Green Monkey kidney 7, and rat osteosarcoma 25/1 mammalian cells. AP activity in these cells, measured three days later, was 40-400-fold above background. When AP and chloramphenicol acetyltransferase (CAT) plasmids were cotransfected, the detection of AP activity by a simple spectrophotometric assay was at least as sensitive as the detection of CAT activity using a radioactive substrate. Moreover, since mammalian AP is a membrane-bound ectoenzyme, transfected cells can be visualized by histochemical staining. This approach was used to estimate transfection efficiency. The convenient methods for AP detection should make it a useful reporter enzyme.

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